



Influence of *Nauclea latifolia* Leaf Extracts on Some Haematological Parameters of Rats on Coconut Oil and Non-coconut Oil Meals

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Authors' contributions

This work was carried out in collaboration between all authors. Authors MIA and HA designed the study and performed the statistical analysis. Authors MIA and UEB wrote the protocol and wrote the first draft of the manuscript. Authors UFU and UEB managed the analyses of the study and the literature searches. All authors read and approved the final manuscript.

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ABSTRACT

Aims: The haematological effects of *Nauclea latifolia* leaf extract on normal mature albino Wistar rats and normal mature hyperlipidemic rats were being investigated.

Study Design: 55 albino rats weighing between 200 – 230 g were divided into 3 experiments. Group 2, 3, 4 of Experiment 1 were administered with 170, 340 and 540 mg/kg body weight of water extract of *Nauclea latifolia* leaves respectively and Group 1 served as control for both experiment 1 and 2. Group 1, 2 and 3 of Experiment 2 received 170, 340 and 510 mg/kg body weight of water soluble ethanol extract while the animals in Experiment 3 were fed with coconut oil meal for 8 weeks before administration of 170, 340 and 510 mg/kg of water soluble fraction ethanol extract to Group 2, 3, 4 respectively while Group 1 served as control. All extracts were administered orally for two weeks.

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Results: Analysis of the haematological indices at $p < 0.05$ showed that there were no significant changes in the WBC counts hence the extracts had no deleterious effect on the defense mechanism of the rats. Similar observations were made for Hb concentration and PCV counts. There was a significant increase in RBC counts in Experiment 1 and 3 and a decreased RBC counts in Experiment 2. Platelet counts showed significant increase in Experiment 1 and 2 but there were no significant changes in Experiment 3, implying that the *Nauclea latifolia* may improve haematological functions in coconut oil induced hyperlipidemia.

Conclusion: This study shows that the leaf extract of *Nauclea latifolia* especially the water extract had an overall beneficiary effect on haematological parameters of albino rats.

Keywords: *Nauclea latifolia*; hyperlipidemia; haematological parameters; coconut oil meal.

1. INTRODUCTION

Traditional plants have been utilized for medicinal purposes for ages by almost all countries of the world [1]. Most plants have been found to possess antidiabetic, antihypertensive, antibacterial and antimalarial properties, etc [2]. This is because plants have myriads of phytochemicals capable of eliciting these effects hence plants used in traditional medicine have served as the source of drug discovery of most modern medicine [3], hence the increasing interest in the pharmacological effects of native plants.

Nauclea latifolia is a shrub commonly called pincushion tree. It is known as mbom-ibong (Ibibio), tabashiya (Hausa), ubuluim (Igbo) and scille maritime (French). It has been reported to possess hypoglycemic property [4] and anti-ulcerogenic activity in ulcer induced albino Wistar rats [5]. It is also used in traditional medication for the treatment of malaria and hypertension as well as diabetes mellitus [6]. The medicinal property of this plant is attributed to the presence of phytochemicals such as alkaloids, saponins, polyphenols which are present in the leaves of this plant [7].

Nauclea latifolia plants have been studied at different levels to evaluate its effects on biological functions. Taiwe et al. [8] investigated the effects of root decoction of *Nauclea latifolia* on the peripheral and central nervous system and its possible mechanisms of action. The decoction was found to have an antipyretic effect [9]. The leaf extract of the plant has also been studied for its antiplasmodial effect [10], antibacterial activities [11,12], as well as antifungal activities [13]. Extensive work has been done on the leaves and root of *Nauclea latifolia*. Furthermore, the fruits have also been studied for its hypocholesterolemic effects [14]. However, the crude extract of the plant material has been reported to be non-toxic at low dosage

but at high dosage and prolonged administration, possess the tendency to adversely affect hepatic function of Chinchilla rabbit [15].

Hyperlipidemia is the condition of excess lipid in the blood. Hyperlipidemic rats are known to have alterations in haematologic profile. Hyperlipidemia in human is associated with atherosclerosis. In light of the increasing interest in the usage of this plant as healing agents in traditional medicine and the availability of limited information on its effect on haematological profile of normal and hyperlipidemic rats. This study was undertaken to assess its effects on haematological indices of normal rats and rats fed with coconut oil meal induced hyperlipidemia so as to evaluate whether the administration of aqueous or ethanolic extract of *Nauclea latifolia* could be useful in the management of haematological alterations in hyperlipidemic rats.

2. MATERIALS AND METHODS

2.1 Animals and Reagents

Fifty-five (55) mature male albino Wistar rats weighing between 200-230 g were obtained from the animal house of the Department of Biochemistry, Faculty of Basic Medical Sciences, University of Calabar, Nigeria. The Animals were kept in standard laboratory condition and handled in accordance with the international guide for the care and use of laboratory animals. All the reagents and chemicals used in this study were of analytical grades.

2.2 Sample Collection and Preparation

Nauclea latifolia leaves were collected from the premises of the Medical School, university of Calabar, Nigeria. The leaves were identified by the Botanist in the Botanical Garden of the University. The leaves were dried in a Plus II oven (Gallenkamp, leicester, UK) at 55°C and crushed using laboratory blender. The ground

leaves were stored in a glass bottle with a plastic screw cap and kept in the refrigerator (4°C). Ethanol extraction of the crushed leaves was carried out by Soxhlet Extraction method and the extract was dried in an oven to remove all the ethanol after which it was stored in the refrigerator until required for use. Water extraction was carried out by soaking chopped fresh leaves in 500 ml distilled water in mortar and thereafter crushed manually by pestle for 2 hours. The residue was filtered using Whatman No.1 filter paper. The concentration was measured by drying a known volume and measuring the dry weight. The ethanol extract was re-extracted in cool water for use and the concentration measured gravimetrically.

2.3 Preparation of Coconut Oil Meal

Coconut oil meal was prepared using the method described by Akpanabiatu et al. [16].

2.4 Experimental Design and Administration

The fifty-five (55) albino rats were divided into eleven (11) groups of five (5) rats per group. The study was carried out in three different experiments;

In Experiment 1, Group 1 (normal control) was subjected to oral administration of 1.5 ml of normal saline as placebo while Group 2, 3 and 4 were treated with 170, 340 and 510 mg/kg body weight respectively of water extract of *Nauclea latifolia* daily for two weeks.

Experiment 2 shared the same control with Experiment 1. Group 1, 2 and 3 were respectively gavaged 170, 340, 510 mg/kg body weight of water soluble fraction of the ethanol extract of *Nauclea latifolia* daily for two weeks.

In Experiment 3, Group 1 (coconut oil meal control) was fed with 10% coconut oil meal diet for 8 weeks before the treatment with normal saline as placebo while Group 2, 3 and 4 were fed coconut oil meal diet for 8 weeks before treatment with 170, 340 and 510 mg/kg body weight respectively of water soluble fraction of the ethanol extract of the leaves for two weeks.

2.5 Determination of Haematological Parameters

At the end of the experiment which lasted for 10 weeks, the experimental animals were

anesthetized under chloroform vapour 24 hours after the last administration. Blood sample was collected by cardiac puncture into EDTA sample bottles. Packed cell volume (PCV), Red Blood Cell (RBC), White Blood Cell (WBC) counts were determined using the method of Dacie and Lewis [17]. Total platelet count determination was carried out on the whole blood by diluting the blood with appropriate fluid and visual observations of the platelet under the microscope. Haemoglobin (Hb) in the whole blood was determined using the method of Alexander and Griffiths [18].

2.6 Statistical Analysis

The data were analyzed using standard student's t-test and $p = .05$ was regarded as significant. The group data are expressed as Mean \pm Standard Deviation.

3. RESULTS AND DISCUSSION

3.1 Results

Tables 1 and 2 show the effects of water extract and water soluble ethanol extract of *Nauclea latifolia* leaves on some haematological parameters of normal rats while Table 3 shows the effect of water soluble ethanol extract on haematological parameters of albino Wistar rats fed with 10% coconut meal for 8 weeks. Data in the three tables showed non-significant changes in the concentration of haemoglobin for the treated animals compared to the control. The same was observed for the PCV as well as WBC counts. However, the RBC count of rats treated with water extract of the leaves increased significantly in the treated groups, likewise the groups fed with 10% coconut oil meal before treatment with water soluble ethanol extract. But the group treated with water soluble ethanol extract without coconut oil meal showed a significant decrease in RBC count. The platelet counts in Table 1 show a dose dependent significant increase when compared to the control. Whereas the platelet counts in Table 2 significantly increased in Group 3 and Group 4 only when compared to the control. However, there was no significant change in platelet count of coconut oil meal fed rats in Table 3 except in Group 3 which showed a significant decrease.

Table 1. Haematological parameters of mature male Wistar albino rats treated with water extract of *Nauclea latifolia* leaves

Parameters	Group 1	Group 2	Group 3	Group 4
Hb (g/dL)	15.57±0.90	12.37±1.63	13.12±2.12	14.7±1.29
PCV (%)	48.78±0.83	49.5±1.5	48.28±1.29	48.25±1.29
RBC count (x10 ⁹ /L)	2628.98±153.34	2954.98±337.62*	2896.17±128.91*	2944.15±246.08*
WBC count (x10 ⁹ /L)	4.925±1.552	4.450±0.473	4.287±0.520	4.172±0.614
Platelet (x10 ⁹ /L)	89.0±10.98	165.0±6.75*	184.75±9.7*	195.25±12.31*

Values = Mean ± SD, * = significance difference at (p < 0.05)

Table 2. Haematological parameters of mature male Wistar albino rats treated with water soluble fraction of ethanol extract of *Nauclea latifolia* leaves

Parameters	Group 1	Group 2	Group 3	Group 4
Hb (g/dL)	15.57±0.90	14.49±1.24	15.12±0.57	15.22±1.20
PCV (%)	48.78±0.83	50.5±2.06	48.25±1.25	48.25±0.8
RBC count (x10 ⁹ /L)	2628.98±153.34	2488.0±81.63*	2201.8±45.91*	2277.55±175.42*
WBC count (x10 ⁹ /L)	4.925±1.552	5.15±0.24	5.96±0.77	5.720±1.22
Platelet (x10 ⁹ /L)	89.0±10.98	104.25±7.58	143.5±14.57*	125.0±4.96*

Values = mean ± SD, * = significance different at (p < 0.05)

Table 3. Haematological parameters of mature male Wistar albino rats fed with 10% coconut meal before treatment with water soluble fraction of ethanol extract of *Nauclea latifolia* leaves

Parameters	Group 1	Group 2	Group 3	Group 4
Hb (g/dL)	15.4±0.7	13.90±1.80	15.55±0.55	14.83±0.21
PCV (%)	50.25±2.68	47.75±4.6	51.79±1.48	48.5 ±1.12
RBC count (x10 ⁹ /L)	1954.36±93.48	2648.72±72.00*	2421.47±66.67*	2280.6±54.31*
WBC count (x10 ⁹ /L)	4.450±0.26	3.787±0.64	4.975±0.23	5.062±0.34
Platelet (x10 ⁹ /L)	105.50±9.13	103.8±7.12	78.5±13.50*	103.5 ±15.86

Values = mean ± SD, * = significance different at (p < 0.05)

3.2 Discussion

Assessment of haematological parameters can be used to determine the extent of deleterious effect of foreign compounds including plant extracts on the blood constituents of the animals [19]. It can also be used to explain blood selected functions of chemicals/plant extract [20]. Furthermore, haematological parameters provide information regarding the status of bone marrow activity and haemolysis [21]. The Hb concentration, WBC and PCV counts of the treated groups in this study did not show any significant change when compared with the controls. Some plant phytochemicals may have the effect of decreasing or increasing the values of some haematological parameters. Decrease in PCV and Hb may be due to reduced erythropoietic activities which may be as a result of inhibition of heme synthesis – mostly often attributed to the presence of phytate. However, some drugs have been shown not to adversely affect haematological and biochemical indices [22,23]. WBC functions as defense machinery in

the eradication of foreign bodies in the system. They are responsible for phagocytosis in the body. The observed insignificant decrease in WBC counts indicates a decrease in the synthesis of WBC in the experimental animals which is the reverse of normal physiologic response following perception of attack from foreign bodies. This implies that the extract did not affect the defense mechanism of the rats.

Platelets play a role in the clotting of blood. Its reduction constitutes an important cause of generalized bleeding. The platelet counts of the experimental animal gavage water extract of *Nauclea latifolia* when compared to the control showed significant difference, but the coconut oil meal fed rats showed no significant changes. The water extract of *Nauclea latifolia* leaves contains some phytochemicals such as saponins, tannins, phytates, alkaloids, flavonoids as well as cyanogenic glycosides alongside Vitamin A and C [24]. It is possible that one or more of these phytochemicals may have stimulated platelet synthesis while the coconut oil

meal seems to have inhibitory effect on platelet synthesis induced by the *Nauclea latifolia* leaves extract. Saponins and tannins from some plant extracts have been reported to have haemostatic activity [25].

The values of RBC count also increased significantly ($p < 0.05$) in the experimental groups of this study. The observed increase in RBC count may be as a result of increase in erythropoietic activity of the animals. Since RBC is synthesized in the bone marrow, this increase may be due to the effect of crude extract on the activity of the bone marrow. Extracts of other plants have also been observed to elicit similar effects on RBC counts [26]. However, the work done by Tende et al. [27] shows that certain extracts do not have the ability to stimulate erythropoietic activity in animals. The active phytochemicals responsible for these effects are not elucidated, hence the need for further research into elucidating and characterizing the particular active ingredient that elicits each effect and the exact mechanism of action.

4. CONCLUSION

We concluded that leaf extract of *Nauclea latifolia* especially the water extract had no detrimental effect on the haematological parameters of normal mature male albino rats and was beneficial in the management of haematological alterations in hyperlipidemic rats.

CONSENT

It is not applicable.

ETHICAL APPROVAL

All authors hereby declare that "Principles of laboratory animal care" (NIH publication No. 85-23, revised 1985) were followed, as well as specific national laws where applicable. All experiments have been examined and approved by the appropriate ethics committee.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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